

- a) obtaining a fluorescently labeled oligonucleotide containing a nucleic acid sequence recognizable by an enzyme that facilitates nucleic acid ligation;
- b) contacting said oligonucleotide with said enzyme that facilitates nucleic acid ligation of said oligonucleotide; and
- c) continuously detecting said ligation reaction by detecting a change in fluorescence intensity as said nucleic acid ligation proceeds.

65. The method of Claim 64, wherein said process comprises a ligase chain reaction.

Recont'd.

66. A method for continuously detecting a specific nucleic acid sequence that is produced during a process that results in amplification of a specific DNA or RNA sequence, said process comprising:

- (a) obtaining a fluorescent labeled oligonucleotide containing a nucleic acid sequence recognized by an enzyme that facilitates nucleic acid cleavage or ligation;
- (b) contacting said oligonucleotide with said enzyme that facilitates nucleic acid ligation or cleavage during a process that results in amplification of said specific DNA or RNA sequence; and
- (c) continuously detecting said ligation or cleavage reaction by detecting a change in fluorescence intensity during said amplification reaction.

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67. A method for continuously detecting a specific nucleic acid sequence by a fluorometric assay comprising the following steps:

(a) obtaining an oligonucleotide that is labeled with a fluorescent acceptor and donor pair and which oligonucleotide is specifically recognized by an enzyme that facilitates nucleic acid ligation or cleavage at a specific nucleic sequence;

(b) contacting said oligonucleotide with said enzyme that facilitates nucleic acid ligation or cleavage of said oligonucleotide; and

(c) continuously detecting said ligation or cleavage reaction by detecting a change in fluorescence intensity.

Cont'd.

68. The method of Claim 67, wherein said continuous detection of said specific nucleic acid sequence occurs while a reaction that results in the amplification of said specific nucleic acid sequence proceeds.

69. The method of Claim 66, wherein said amplification reaction is a catalytic hybridization amplification reaction.

70. The method of Claim 66, wherein said amplification reaction is a polymerase chain reaction.

71. The method of Claim 66, wherein said amplification reaction is a ligase chain reaction.

72. The method of Claim 66, wherein said change in fluorescence intensity is measured by time-resolved fluorescence.

73. The method of Claim 66, wherein said change in fluorescence intensity is measured by energy transfer.

74. The method of Claim 66, wherein said change in fluorescence intensity is measured by single photon counting.

Revised.
75. The method of Claim 66, wherein said change in fluorescence is measured by an analog method.

76. The method of Claim 66, wherein said change in fluorescence is measured by fluorescent lifetime.

77. The method of Claim 67, wherein said fluorescent donor and acceptor are spaced with an about zero and twenty bases of one another.

78. The method of Claim 67, wherein said fluorescent donor and acceptor are spaced within about zero to seven bases of one another.

79. The method of Claim 67, wherein either or both of said fluorescent donor and acceptor are attached to said oligonucleotide by a linker.

80. The method of Claim 79, wherein said linker is a carbon linker comprising a 12 carbon chain.

81. The method of Claim 67, wherein said donor and acceptor are at opposite ends of said specific nucleic acid sequence.

Revised.
82. The method of Claim 67, wherein either said donor or acceptor are attached internally to said specific nucleic acid sequence.

83. The method of Claim 67, wherein said fluorescent donor and acceptor are on different strands of said oligonucleotide.

84. The method of Claim 66, wherein said detected nucleic acid sequence is a DNA.

85. The method of Claim 66, wherein said detected nucleic acid sequence is a double stranded DNA.

86. The method of Claim 66, wherein said detected nucleic acid sequence is an RNA.

87. The method of Claim 67, wherein said detected nucleic acid sequence is a DNA.

Revised.
88. The method of Claim 67, wherein said detected nucleic acid sequence is a double stranded DNA.

89. The method of Claim 67, wherein said detected nucleic acid sequence is an RNA.--

REMARKS

Entry of the foregoing amendments, reconsideration and reexamination of the subject application, as amended, pursuant to and consistent with 37 C.F.R. §1.112, and in light of the remarks which follow, are respectfully requested.